

What is claimed is:

1. Pluripotent embryonic stem cells isolated from *in vitro* treatment of canine embryos.
2. A purified cell preparation comprising cells exhibiting a canine embryonic stem cell phenotype.
- 5 3. A purified cell preparation comprised or enriched for canine embryonic stem cells that are capable of indefinite proliferation *in vitro* in an undifferentiated state.
4. A purified cell preparation as claimed in any preceding claim further characterized by being immunoreactive with markers for canine embryonic stem cells.
5. A purified cell preparation as claimed in claim 4 wherein the markers are not found in murine
10 embryonic stem cells.
6. A purified cell preparation as claimed in claim 4 wherein the markers are alkaline phosphatase, AP, stage-specific embryonic antigen-4 (SSEA-4), TRA-1-60, and Oct4 transcription factor.
7. A purified cell preparation as claimed in any preceding claim induced to differentiate into cells of various lineages.
- 15 8. Cells differentiated *in vitro* from cells of a cell preparation according to any preceding claim.
9. A cell line comprising canine embryonic stem cells, or cells differentiated or derived therefrom.
10. Embryonic stem cells or a cell preparation of any preceding claim wherein the cells are genetically modified.
11. A method for producing purified canine embryonic stem cells comprising the step of culturing inner
20 cell mass cells from a canine embryo under conditions to promote proliferation of undifferentiated cells.
12. A method as claimed in claim 11 which further comprises isolating a canine embryo, culturing the embryo in the presence of a feeder layer and one or more proliferation agent, removing a blastocyst outgrowth and culturing the outgrowth in the presence of a fresh feeder layer.
- 25 13. A method of claim 11 further comprising: (a) obtaining a canine embryo at a morula to blastocyst stage; (b) removing inner cell mass (ICM) cells from the canine embryo; (c) culturing inner cell mass (ICM) cells in the presence of a feeder layer and one or more proliferation agent to promote proliferation of undifferentiated stem cells; and (c) recovering stem cells.
14. A method according to any preceding claim, further comprising passaging the stem cells to prevent
30 differentiation of the cells and to maintain a cell line in culture.
15. A method for producing cells exhibiting a canine embryonic stem cell phenotype comprising (a) obtaining a canine embryo at a morula to blastocyst stage; (b) culturing inner cell mass (ICM) cells from the canine embryo under conditions which promote proliferation of undifferentiated stem cells; and (c) recovering stem cells.
- 35 16. A method of any preceding claim comprising inducing differentiation of the embryonic stem cells into cells that exhibit morphological, physiological, functional, and/or immunological features of somatic and germ cells.
17. A cell preparation or cell line derived from cells cultured in accordance with a method of any preceding claim.

18. Canine transgenic cells, cell lines, or tissues produced using the canine embryonic stem cells of any preceding claim.
19. A blastocyst to which has been introduced one or more canine embryonic stem cells of a preparation of any preceding claim.
- 5 20. An embryonic cell to which has been introduced by nuclear transfer a nucleus of an embryonic stem cell of a preparation of any preceding claim.
21. A chimeric non-human animal which is the progeny of a blastocyst according to claim 19 or embryonic cell of claim 20.
- 10 22. A method which comprises introducing into a blastocyst one or more stem cells made according to a method of any preceding claim.
23. A method which comprises introducing by nuclear transfer into an embryonic cell a nucleus of a stem cell according to claim 1.
24. A method which comprises introducing to the uterus of a pseudo-pregnant foster mother animal a viable embryo obtained using a technique involving a method of any preceding claim.
- 15 25. Use of embryonic stem cells or cell preparations of any preceding claim in the preparation of a medicament for cell and gene therapies aimed at alleviating disease.
26. A use of claim 25 wherein the embryonic stem cells or cell preparations are transplanted in, or grafted to a subject in need.
- 20 27. Use of embryonic stem cells or cell preparations of any preceding claim in the preparation of a medicament for the replacement of body tissues, organs, components or structures which are missing or damaged due to trauma, age, metabolic or toxic injury, disease, or idiopathic loss.
28. A pharmaceutical composition comprising embryonic stem cells or cell preparations of any preceding claim and a pharmaceutically acceptable carrier, excipient, or diluent.
- 25 29. A method for testing toxicity of a drug comprising culturing embryonic stem cells, cell preparations, or cell lines as claimed in any preceding claim in a suitable medium, introducing a drug to the culture and examining cells to determine if the drug has had an adverse effect on the cells.
- 30 30. A method of screening for potential drugs that modulate canine development comprising incubating a test compound and embryonic stem cells as claimed in any preceding claim under conditions sufficient to allow the test compound and stem cells to interact, and determining the effect of the compound on a function of the stem cells before and after incubating with the test compound.
31. A method for assaying the activity of a test substance comprising the steps of:
- 35 (a) exposing embryonic stem cells, cell preparations or cell lines as claimed in any preceding claim to a test substance; and
- (b) detecting the presence or absence of an effect of the test substance on the survival of the cells or on a morphological, functional, or physiological characteristic and/or molecular biological property of the cells, whereby an effect altering cell survival, a morphological, functional, or physiological characteristic and/or a molecular biological property of the cells indicates the activity of the test substance.

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32. A method for screening a potential new drug to treat a disorder comprising the steps of:
- (a) exposing embryonic stem cells, cell preparations or cell lines as claimed in any preceding claim to a potential new drug; and
- 5 (b) detecting the presence or absence of an effect of the potential new drug on the survival of the cells or on a morphological, functional, or physiological characteristic and/or molecular biological property of said cells, whereby an effect altering cell survival, a morphological, functional, or physiological characteristic and/or a molecular biological property of the cells indicates the activity of the potential new drug.
33. A method of treating a condition or disease in a subject comprising administering an effective amount of embryonic stem cells or cell preparations as claimed in any preceding claim.
- 10 34. A method of claim 33 wherein the condition or disease is hemophilia, muscular dystrophy, MPS-1, glycogen storage disease, narcolepsy, thrombasthenia, Von Willebrand Disease, osteogenesis, nephritis, retinal atrophy, severe combined immunodeficiency disease, hematopoietic disorder, autoimmune disorder, cancer, heart disease, motor neuron disease, degenerative bone and joint diseases, and atherosclerosis.
- 15 35. A method for conducting a stem cell business comprising (a) identifying one or more agents which affect the proliferation, differentiation, function, or survival of embryonic stem cells, cell preparations, or cell lines of any preceding claim; (b) conducting therapeutic profiling of agents identified in (a); or analogs thereof for efficacy and toxicity in animals; and (c) formulating a pharmaceutical composition including one or more agents identified in (b) as having an acceptable therapeutic profile.
- 20 36. A kit comprising embryonic stem cells as claimed in any preceding claim and instructions for their use.
37. A primer that hybridizes to an Oct4 canine nucleotide sequence.
- 25 38. A primer comprising the sequence of SEQ ID No. 1, 2, 5 or 6.